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L68 ANSWER 1 OF 21 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
    DUPLICATE 1
                    2004:261058 BIOSIS
ACCESSION NUMBER:
DOCUMENT NUMBER:
                    PREV200400262827
TITLE:
                    Large-scale separation and production of engineered
                    proteins, designed for facilitated recovery in
                    detergent-based aqueous two-
                    phase extraction systems.
AUTHOR (S):
                    Selber, Klaus; Tjerneld, Folke; Collen, Anna; Hyytia,
                    Teppo; Nakari-Setala, Tiina; Bailey, Michael; Fagerstrom,
                    Richard; Kan, John; van der Laan, Joop; Penttila, Merja;
                    Kula, Maria-Regina [Reprint Author]
                    Institute of Enzyme Technology, Heinrich-Heine University,
CORPORATE SOURCE:
                    Juelich, Duesseldorf, D-52426, Germany
                    mrk3372002@yahoo.de
SOURCE:
                    Process Biochemistry, (March 31 2004) Vol. 39, No. 7, pp.
                    889-896. print.
                    ISSN: 1359-5113 (ISSN print).
DOCUMENT TYPE:
                    Article
LANGUAGE:
                    English
ENTRY DATE:
                    Entered STN: 19 May 2004
                    Last Updated on STN: 19 May 2004
     The feasibility and scalability of extraction in detergent-based
AB
     aqueous two-phase systems for the separation
     of proteins from culture broth is demonstrated. At the same time the
     large-scale production of a fusion protein and the influence of
     cultivation scale on the efficiency of separation were investigated. An
     amphiphilic fusion protein EGIcore-HFBI was chosen, consisting
     of the catalytic core of the cellulase endoglucanase I and the small
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protein hydrophobin I, expressed homologously in Trichoderma

reesei. Using the technical nonionic detergent Agrimul NRE 1205 the separation was successfully scaled up to 1200 1. No differences in yield or in partition coefficient were observed at 10 ml and 1200 1 scale. Changes in the fermentation temperature and scale, however, can influence the properties of the protein and thus alter partition coefficient and yield. The decreased separation efficiency appears to be correlated with changes in glycosylation at lower cultivation temperatures.

L68 ANSWER 2 OF 21 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN

ACCESSION NUMBER: 2004

2004:324471 SCISEARCH

THE GENUINE ARTICLE: 806GQ

TITLE: Large-scale separation and production of engineered

proteins, designed for facilitated recovery in

detergent-based aqueous twophase extraction systems

AUTHOR: Selber K; Tjerneld F; Collen A; Hyytia T; Nakari-Setala T;

Bailey M; Fagerstrom R; Kan J; van der Laan J; Penttila M;

Kula M R (Reprint)

CORPORATE SOURCE: Univ Dusseldorf, Inst Enzyme Technol, D-52426 Dusseldorf,

Juelich, Germany (Reprint); Lund Univ, Ctr Chem & Chem Engn, Dept Biochem, S-22100 Lund, Sweden; VTT Biotechnol & Food Rog, FIN 02044 Egges, Finland, Germany Lat. RV

Food Res, FIN-02044 Espoo, Finland; Genencor Int BV,

NL-2333 CN Leiden, Netherlands

COUNTRY OF AUTHOR: / Germany; Sweden; Finland; Netherlands

SOURCE: PROCESS BIG

PROCESS BIOCHEMISTRY, (31 MAR 2004) Vol. 39, No. 7, pp.

889-896.

Publisher: ELSEVIER SCI LTD, THE BOULEVARD, LANGFORD LANE,

KIDLINGTON, OXFORD OX5 1GB, OXON, ENGLAND.

ISSN: 0032-9592.

DOCUMENT TYPE:

ΔR

Article; Journal

LANGUAGE: REFERENCE COUNT:

PATENT INFORMATION:

APPLICATION INFO.:

English

\*ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS\*

The feasibility and scalability of extraction in detergent-based aqueous two-phase systems for the separation of proteins from culture broth is demonstrated. At the same time the

of proteins from culture broth is demonstrated. At the same time the large-scale production of a fusion protein and the influence of cultivation scale on the efficiency of separation were investigated. An amphiphilic fusion protein EGIcore-HFBI was chosen, consisting of the catalytic core of the cellulase endoglucanase I and the small protein hydrophobin I, expressed homologously in Trichoderma reesei. Using the technical nonionic detergent Agrimul NRE 1205 the separation was successfully scaled up to 1200 l. No differences in yield or in partition coefficient were observed at 10 ml and 1200 l scale. Changes in the fermentation temperature and scale, however, can influence the properties of the protein and thus alter partition coefficient and yield. The decreased separation efficiency appears to be correlated with changes in glycosylation at lower cultivation temperatures. (C) 2003 Published by Elsevier Ltd.

L68 ANSWER 3 OF 21 USPATFULL on STN

ACCESSION NUMBER: 2002:241975 USPATFULL

TITLE: Electrophoretic medium

Electrophoretic medium and process for the production

thereof

INVENTOR(S): Paolini, Richard J., JR., Arlington, MA, UNITED STATES

Miller, David D., Billerica, MA, UNITED STATES Comiskey, Barrett, Cambridge, MA, UNITED STATES

NUMBER KIND DATE
----US 2002131147 A1 20020919
US 2002-683903 A1 20020228 (9)

NUMBER DATE

PRIORITY INFORMATION: US 2001-277079P 20010319 (60)

US 2001-277391P 20010319 (60)

DOCUMENT TYPE: Utility
FILE SEGMENT: APPLICATION

LEGAL REPRESENTATIVE: DAVID J COLE, E INK CORPORATION, 733 CONCORD AVE,

CAMBRIDGE, MA, 02138-1002

NUMBER OF CLAIMS: 29 EXEMPLARY CLAIM: 1

NUMBER OF DRAWINGS: 2 Drawing Page(s)

LINE COUNT: 1244

A two-phase electrophoretic medium comprises a continuous phase and a discontinuous phase. The discontinuous phase comprises a plurality of droplets, each of which comprises a suspending fluid and at least one particle disposed within the suspending fluid and capable of moving through the fluid upon application of an electric field to the electrophoretic medium. The continuous phase surrounds and encapsulates the discontinuous phase. The discontinuous phase comprises at least about 40 percent by volume of the electrophoretic medium.

L68 ANSWER 4 OF 21 AGRICOLA Compiled and distributed by the National Agricultural Library of the Department of Agriculture of the United States of America. It contains copyrighted materials. All rights reserved.

(2004) on STN DUPLICATE 2

ACCESSION NUMBER: 2003:1946 AGRICOLA

DOCUMENT NUMBER: IND23297051

TITLE: Expression of a fungal hydrophobin in the

Saccharomyces cerevisiae cell wall: effect on cell

surface properties and immobilization.

Nakari-Setala, T.; Azeredo, J.; Henriques, M.;

Oliveira, R.; Teixeira, J.; Linder, M.; Penttila, M.

Applied and environmental microbiology, July 2002.

Vol. 68, No. 7. p. 3385-3391

Publisher: Washington : American Society for

Microbiology

CODEN: AEMIDF; ISSN: 0099-2240

OTE: Includes references

District of Columbia; United States

DOCUMENT TYPE: Article

AUTHOR (S):

PUB. COUNTRY:

SOURCE:

FILE SEGMENT: U.S. Imprints not USDA, Experiment or Extension

LANGUAGE: English
AB The aim of this work was to

The aim of this work was to modify the cell surface properties of Saccharomyces cerevisiae by expression of the HFBI hydrophobin of the filamentous fungus Trichoderma reesei on the yeast cell surface. The second aim was to study the immobilization capacity of the modified cells. Fusion to the Flo1p flocculin was used to target the HFBI moiety to the cell wall. Determination of cell surface characteristics with contact angle and zeta potential measurements indicated that HFBI-producing cells are more apolar and slightly less negatively charged than the parent cells. Adsorption of the yeast cells to different commercial supports was studied. A twofold increase in the binding affinity of the hydrophobin-producing yeast to hydrophobic silicone-based materials was observed, while no improvement in the interaction with hydrophilic carriers could be seen compared to that of the parent cells. Hydrophobic interactions between the yeast cells and the support are suggested to play a major role in attachment. Also, a slight increase in the initial adsorption rate of the hydrophobin yeast was observed. Furthermore, due to the engineered cell surface, hydrophobin-producing yeast cells were efficiently separated in an aqueous two-phase system by using a nonionic polyoxyethylene detergent, C(12-18)EO(5).

L68 ANSWER 5 OF 21 CEABA-VTB COPYRIGHT 2004 DECHEMA on STN ACCESSION NUMBER: 2002(10):5428 CEABA-VTB FILE SEGMENT TITLE: Primary recovery of a genetically engineer of the primary recovery of the prima

2002(10):5428 CEABA-VTB FILE SEGMENT B Primary recovery of a genetically engineered Trichoderma reesei endoglucanase I (Cel 7B) fusion protein in cloud point extraction systems AUTHOR: Collen, A.; Selber, K.; Hyytiae, T.; Persson, J.;

Nakari-Setlae, T.; Bailey, M.; Fagerstroem, R.; Kula, M.-R.; Penttilae, M.; Staalbrand, H.; Tjerneld, F.

Lund Univ., S; VTT Biotechnol., Espoo, FIN; Univ.

Duesseldorf, Juelich, D

SOURCE: Biotechnology and Bioengineering (2002) 78(4), 51

Reference(s), 385-394, 4f, 3t CODEN: BIBIAU ISSN: 0006-3592

DOCUMENT TYPE: Journal LANGUAGE: English

FS B

CORPORATE SOURCE:

AB

SOURCE:

to

PUBLISHER:

Aqueous two-phase extraction or cloud point

extraction systems (CPE) is designated as detergent based system used to separate hydrophobic from hydrophilic proteins to increase the specificity of such systems affinity derived surfactants have been employed and a hydrophilic cellulose from Trichoderma reesei called endogluccanase I (EGI) was partitioned to thin the detergent-based system by the fusion of a hydrophobic protein (hydrophobin) to the target protein. Here the partitioning of hydrophilic EGI by fusion of peptide tags to the protein is studied and the expression of fused protein under large scale conditions was studied. The partitioning of the T. reesei strain (WP4) tag was shown to strongly enhance partitioning of the tagged protein to the detergent-enriched phase leaving unmodified cellulases and bulk proteins in the water phase (Steen, Helen J.)

L68 ANSWER 6 OF 21 CAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 2002:766622 CAPLUS

DOCUMENT NUMBER: 137:368622

TITLE: Parameters influencing protein extraction for whole

broths in detergent based aqueous

two-phase systems

AUTHOR(S): Selber, Klaus; Collen, Anna; Hyytia, Teppo; Penttila,

Merja; Tjerneld, Folke; Kula, Maria-Regina

CORPORATE SOURCE: Institut fur Enzymtechnologie, Heinrich-Heine-

Universitat Dusseldorf, Julich, D-52426, Germany

Bioseparation (2002), Volume Date 2001, 10(4/5),

229-236

CODEN: BISPE4; ISSN: 0923-179X

Kluwer Academic Publishers

DOCUMENT TYPE: Journal LANGUAGE: English

The parameters important for an optimization of cloud point extraction in tech. scale were investigated using a genetically engineered fusion protein derived from endoglucanase I expressed in Trichoderma reesei and the nonionic polyoxyethylene Agrimul NRE 1205. The key parameters are temperature, detergent concentration, and addnl. salts. These parameters are interdependent, thus there is an optimum in the partition coefficient with respect to detergent

concentration and a maximum for the partition coefficient and the yield with respect

temperature These results were confirmed for the detergent C12E5 to demonstrate that these optima are due to the nature of polyoxyethylenes. Cloud point extraction was found to be only slightly affected by pH. In the case studied extraction of whole broth is favorable for a high yield and partition coefficient, since fusion protein adhering to the cells can be solubilized. However

some loss of detergent which remains in the fungal biomass was observed
REFERENCE COUNT: 34 THERE ARE 34 CITED REFERENCES AVAILABLE FOR THIS

RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L68 ANSWER 7 OF 21 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN

DUPLICATE 3

ACCESSION NUMBER: 2002:260341 BIOSIS

DOCUMENT NUMBER: PREV200200260341

TITLE: A novel two-step extraction method with detergent/polymer

systems for primary recovery of the fusion protein

endoglucanase I-hydrophobin I.

AUTHOR(S): Collen, Anna; Persson, Josefine; Linder, Markus;

Nakari-Setala, Tiina; Penttila, Merja; Tjerneld, Folke

[Reprint author]; Sivars, Ulf

Department of Biochemistry, Center for Chemistry and ORPORATE SOURCE:

Chemical Engineering, Lund University, S-221 00, Lund,

folke.tjerneld@biokem.lu.se

Biochimica et Biophysica Acta, (15 January, 2002) Vol. OURCE:

1569, No. 1-3, pp. 139-150. print.

CODEN: BBACAQ. ISSN: 0006-3002.

OCUMENT TYPE:

Article English

ANGUAGE: NTRY DATE:

В

Entered STN: 24 Apr 2002

Last Updated on STN: 24 Apr 2002

Extraction systems for hydrophobically tagged proteins have been developed based on phase separation in aqueous solutions of non-ionic detergents and polymers. The systems have earlier only been applied for separation of membrane proteins. Here, we examine the partitioning and purification of

the amphiphilic fusion protein endoglucanase Icore-hydrophobin I

(EGIcore-HFBI) from culture filtrate originating from a

Trichoderma reesei fermentation. The micelle extraction system was formed by mixing the non-ionic detergent Triton X-114 or Triton X-100 with the hydroxypropyl starch polymer, Reppal PES100. The detergent/polymer aqueous two-phase systems resulted in both

better separation characteristics and increased robustness compared to cloud point extraction in a Triton X-114/water system. Separation and robustness were characterized for the parameters: temperature, protein and salt additions. In the Triton X-114/Reppal PES100 detergent/polymer  ${\tt system} \ {\tt EGIcore-{\bf HFBI}} \ {\tt strongly} \ {\tt partitioned} \ {\tt into} \ {\tt the} \ {\tt micelle-rich}$ phase with a partition coefficient (K) of 15 and was separated from

hydrophilic proteins, which preferably partitioned to the polymer phase. After the primary recovery step, EGIcore-HFBI was quantitatively back-extracted (KEGIcore-HFBI=150, yield=99%) into a water

In this second step, ethylene oxide-propylene oxide (EOPO) copolymers were added to the micelle-rich phase and temperature-induced phase separation at 55degreeC was performed. Total recovery of EGIcore-HFBI after the two separation steps was 90% with a volume

reduction of six times. For thermolabile proteins, the back-extraction temperature could be decreased to room temperature by using a hydrophobically modified EOPO copolymer, with slightly lower yield. addition of thermoseparating co-polymer is a novel approach to remove

detergent and effectively releases the fusion protein EGIcore-HFBI into a water phase.

68 ANSWER 8 OF 21 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN

CCESSION NUMBER: 2002:287918 SCISEARCH

HE GENUINE ARTICLE: 536GM

ITLE: A novel two-step extraction method with detergent/polymer

systems for primary recovery of the fusion protein

endoglucanase I-hydrophobin I

UTHOR: Collen A; Persson J; Linder M; Nakari-Setala T; Penttila

M; Tjerneld F (Reprint); Sivars U

ORPORATE SOURCE: Lund Univ, Ctr Chem & Chem Engn, Dept Biochem, POB 124,

S-22100 Lund, Sweden (Reprint); Lund Univ, Ctr Chem & Chem Engn, Dept Biochem, S-22100 Lund, Sweden; VTT Biotechnol &

Food Res, FIN-02044 Espoo, Finland

OUNTRY OF AUTHOR: Sweden; Finland

OURCE: BIOCHIMICA ET BIOPHYSICA ACTA-GENERAL SUBJECTS, (15 JAN

2002) Vol. 1569, No. 1-3, pp. 139-150.

Publisher: ELSEVIER SCIENCE BV, PO BOX 211, 1000 AE

AMSTERDAM, NETHERLANDS.

ISSN: 0304-4165. Article; Journal

OCUMENT TYPE: ANGUAGE:

English

**EFERENCE COUNT:** 

\*ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS\*

Extraction systems for hydrophobically tagged proteins have been

developed based on phase separation in aqueous solutions of non-ionic detergents and polymers. The systems have earlier only been; applied for separation of membrane proteins. Here, we examine the partitioning and purification of the amphiphilic fusion protein endoglucanase I-core-hydrophobin I (EGI(core)-HFBI) from culture filtrate originating from a Trichoderma reesei fermentation. The micelle extraction system was formed by mixing the non-ionic detergent Triton X-114 or Triton X-100 with the hydroxypropyl starch polymer, Reppal PES100. The detergent/polymer aqueous twophase systems resulted in both better separation characteristics and increased robustness compared to cloud point extraction in a Triton X-114/water system. Separation and robustness were characterized for the parameters: temperature, protein and salt additions. In the Triton X-114/Reppal PES100 detergent/polymer system EGI(core)-HFBI strongly partitioned into the micelle-rich phase with a partition coefficient (K) of 15 and was separated from hydrophilic proteins, which preferably partitioned to the polymer phase. After the primary recovery step, EGI(core)-HFBI was quantitatively back-extracted (KEGIcore-HFBI = 150, yield = 99%) into a water phase. In this second step, ethylene oxide-propylene oxide (EOPO) copolymers were added to the micelle-rich phase and temperature-induced phase separation at 55degreesC was performed. Total recovery of EGI(core)-HFBI after the two separation steps was 90% with a volume reduction of six times. For thermolabile proteins, the back-extraction temperature could be decreased to room temperature by using a hydrophobically modified EOPO copolymer, with slightly lower yield. The addition of thermoseparating co-polymer is a novel approach to remove detergent and effectively releases the fusion protein EGI(core)-HFBI into a water phase. (C) 2002 Elsevier Science B.V. All rights reserved.

L68 ANSWER 9 OF 21 ANABSTR COPYRIGHT 2004 RSC on STN DUPLICATE 4  $\label{eq:endoglucanases} \textbf{Emdoglucanases (EGI) (endo-1,4-$\beta-$D-$glucan-$4-$glucanohydrolase, EC} \\$ 3.2.1.4, Ce17B) of Trichoderma reesei are industrially important enzymes. Thus, there is a great need for development of a primary recovery method suitable for large-scale utilization. In this study we present a concept applicable for large-scale purification of an EGI fusion protein by one-step extraction in a poly(ethylene glycol) PEG-sodium/potassium phosphate aqueous two-phase system. EGI is a two-module enzyme composed of an N-terminal catalytic module and a C-terminal cellulose binding module (CBM) separated by a glycosylated linker region. Partitioning of six different EGI constructs, containing the C-terminal extensions (WP)2, (WP)4 or the amphiphilic protein hydrophobin I (HFB) of T. reesei instead of the CBM were studied to evaluate if any of the fusions could improve the partition coefficient sufficiently to be suitable for large-scale production. All constructs showed improved partitioning in comparison to full length EGI. The (WP)4 extensions resulted in 26- to 60-fold improvement of partition coefficient. Consequently, a relative minor change in amino-acid sequence on the two-module protein EGI improved the partition coefficient significantly in the PEG 4000-sodium/potassium phosphate system. The addition of **HFBI** to EGI clearly enhanced the partition coefficient (K = 1.2) in comparison to full-length EGI (K = 0.035). Partitioning of the construct with (WP)4 fused to the catalytic module and a short sequence of the linker (EGIcore-P5(WP)4) resulted in the highest partition coefficient (K = 54) and a yield of 98% in the PEG phase. Gel electrophoresis showed that the construct with the (WP)4 tag attached after a penta-proline linker could be purified from the other bulk proteins by only a single-step separation in the PEG 4000-sodium/potassium phosphate system. This is a major improvement in comparison with the previously studied model (ethylene oxide-propylene oxide)-dextran system. Hence, this construct will be suitable for further optimization of the extraction of the enzyme in a PEG 4000-sodium/potassium phosphate system from culture filtrate.

ANSWER 10 OF 21 DISSABS COPYRIGHT (C) 2004 ProQuest Information and Learning Company; All Rights Reserved on STN

ACCESSION NUMBER: 2002:9380 DISSABS Order Number: AAIC806246 (not

available for sale by UMI)

TITLE: Hydrophobic fusion tags: Implication for bioseparation and

cellular expression

Collen, Anna Maria Christine [Ph.D.] AUTHOR: CORPORATE SOURCE: Lunds Universitet (Sweden) (0899)

SOURCE: Dissertation Abstracts International, (2001) Vol. 62, No.

4C, p. 565. Order No.: AAIC806246 (not available for sale

by UMI).

ISBN: 91-7874-140-8.

OCUMENT TYPE: Dissertation

TILE SEGMENT: DAT LANGUAGE: English

> The studies in this thesis have shown that the partitioning of endoglucanase I (EGI, Cel7B) from Trichoderma reesei could be significantly improved by relatively minor genetic engineering. By adding short peptides composed of tryptophan and proline residues to EGI, extreme partitioning could be obtained. The site of the tag fission was shown to

be crucial for the efficiency of the tag. Methods suitable for large-scale purification of genetically modified EGI by a single-step extraction in aqueous two-phase systems have been

established. The most optimal fusion protein, with respect to partitioning enhancement resulted, however, in impaired production in T. reesei. This was further elucidated and the low production was suggested to be caused by several factors such as proteolysis, impaired secretion and intracellular accumulation of the hydrophobic fusion protein. At certain stages during growth of the transformant expressing EGIcore-p5(WP)4 slight induction of the gene encoding the ER residual chaperone BIPI was detected.

Furthermore, the amphiphilic protein hydrophobin I was utilized as a fusion tag to direct partitioning in aqueous two phase systems. A system with improved separation features was evaluated, which is a clear progression from previously used systems with respect to both robustness and purification properties. Applications towards large-scale purification with this system might be possible in the foreseeable future. Additionally, a novel approach for detergent removal after two-phase extraction in detergent based systems was developed. By addition of thermoseparating polymers, HM-EOPO or EOPO, phase separation could be induced by temperature increase, and thus the fusion protein could be recovered in a water phase. This method is both environmentally benign and displays compatibility with subsequent purification steps and handling of the target protein.

68 ANSWER 11 OF 21 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN DUPLICATE 5

2002:267630 BIOSIS CCESSION NUMBER: OCUMENT NUMBER: PREV200200267630

'ITLE: The hydrophobins HFBI and HFBII

from Trichoderma reesei showing efficient interactions with

nonionic surfactants in aqueous two-

phase systems.

UTHOR (S): Linder, Markus [Reprint author]; Selber, Klaus;

Nakari-Setala, Tiina; Qiao, Mingqiang; Kula, Maria-Regina;

Penttila, Merja

ORPORATE SOURCE: VTT Biotechnology, FIN-02044, Espoo, Finland

markus.linder@vtt.fi

OURCE: Biomacromolecules, (Summer, 2001) Vol. 2, No. 2, pp.

511-517. print.

ISSN: 1525-7797.

Article

OCUMENT TYPE: ANGUAGE: English NTRY DATE:

В

Entered STN: 1 May 2002

Last Updated on STN: 1 May 2002

Fungal hydrophobins are a group of surface active, self-assembling proteins. The filamentous fungus Trichoderma reesei produces two (class II) hydrophobins, HFBI and We have studied how these water-soluble hydrophobins behave in two-phase systems using a series of nonionic surfactants with different characteristics. It was found that both hydrophobins, but especially HFBI, had a very high affinity for the surfactants. The highest partitioning coefficient, over 2500, was observed for HFBI with C11EO2. Reducing the disulfides in the protein resulted in a complete loss of affinity for the surfactant, which demonstrates that the interaction is dependent on the disulfide-stabilized conformation. The hydrophobins could be efficiently extracted back from the surfactant phase by addition of alcohols such as isobutanol. Effects of the type of surfactant, temperature, pH, and ionic strength were investigated. The use of this method for purifying the proteins from crude fungal culture supernatants is demonstrated and implications of the protein-polymer interaction are discussed.

L68 ANSWER 12 OF 21 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

ACCESSION NUMBER: 2002-10300 BIOTECHDS TITLE: Parameters influencing

Parameters influencing protein extraction for whole broths in detergent based agreeus two-phase

detergent based aqueous two-phase

systems;

vector-mediated fusion gene transfer, expression in Trichoderma reesei and mathematical model for recombinant

protein production and downstream processing

AUTHOR: SELBER K; COLLEN A; HYYTIA T; PENTTILA M; TJERNELD F; KULA MR

CORPORATE SOURCE: Univ Dusseldorf; Lund Univ; VTT Biotechnol

LOCATION: Selber K,

AΒ

SOURCE: BIOSEPARATION; (2001) 10, 4-5, 229-236

ISSN: 0923-179X

DOCUMENT TYPE: Journal LANGUAGE: English AN 2002-10300 BIOTECHDS

AUTHOR ABSTRACT - The parameters important for an optimisation of cloud point extraction in technical scale were investigated using a genetically engineered fusion protein derived from endoglucanase I expressed in Trichoderma reesei and the nonionic polyoxyethylene Agrimul NRE 1205. The key parameters are temperature, detergent concentration, and additional salts. These parameters are interdependent, thus there is an optimum in the partition coefficient with respect to detergent concentration and a maximum for the partition coefficient and the yield with respect to temperature. These results were confirmed forthe detergent C12E5 to demonstrate that these optima are due to the natureof polyoxyethylenes. Cloud point extraction was found to be only slightly affected by pH. In the case studied extraction of whole broth is favourablefor a high yield and partition coefficient, since fusion protein adhering to the cells can be solubilized. However some loss of detergent which remains in the fungal biomass was observed. (8 pages)

L68 ANSWER 13 OF 21 CAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER: 2001:197443 CAPLUS

TITLE: Protein extraction using aqueous two

-phase systems

AUTHOR(S): Kula, Maria-Regina A.; Selber, Klaus

Institute of Enzyme Technology, Heinrich Heine

University Duesseldorf, D-52428 Juelich, Germany

Abstracts of Papers - American Chemical Society

(2001), 221st, BIOT-136

CODEN: ACSRAL; ISSN: 0065-7727 American Chemical Society

PUBLISHER: American Chemical Society
DOCUMENT TYPE: Journal; Meeting Abstract

LANGUAGE: English

CORPORATE SOURCE:

SOURCE:

AB Solvent extraction is widely used in industry to isolate labile compds.

Proteins are amenable to extraction using aq. two-

phase systems. This approach has the advantage, that whole broth

or cell homogenates can be processed integrating product capture with the

removal of solids in a single unit operation. Continuous isolation of enzymes from homogenates will be demonstrated in pilot scale processing 500 kg yeast per day. The very low interfacial tension of these systems allows mixing with low energy input and fast approach to equilibrium, which has to be taken into special consideration when operating conventional multistage extraction equipment. The interfacial tension may limit centrifugal phase separation The latter was observed during the isolation of an endoglucanase I- hydrophobin fusion protein from Trichoderma spec.in a cloud point extraction Gravity settling is a useful, low cost option to sep. aqueous phases since emulsions are rarely encountered.

L68 ANSWER 14 OF 21 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN

ACCESSION NUMBER: 2001-03035 BIOTECHDS

TITLE: Isolation and purification of proteins or cells in

aqueous two-phase systems,

comprises combining a desired protein or a cell to a targeting protein capable of isolating the protein or cell

into one of the phases;

vector-mediated CM-cellulase, hemicellulase, protease or antibody gene transfer and expression in Saccharomyces cerevisiae for recombinant protein production and protein

purification

AUTHOR: Penttila M; Nakari-Setala T; Fagerstrom R; Selber K; Kula M

R; Linder M; Tjerneld F

PATENT ASSIGNEE: VTT

LOCATION: Espoo, Finland.

PATENT INFO: WO 2000058342 5 Oct 2000 APPLICATION INFO: WO 2000-FI249 24 Mar 2000

PRIORITY INFO: FI 1999-1782 20 Aug 1999; FI 1999-667 25 Mar 1999

DOCUMENT TYPE: Patent LANGUAGE: English

ΑN

AB

INVENTOR(S):

PATENT ASSIGNEE(S):

OTHER SOURCE: WPI: 2000-686858 [67]

2001-03035 BIOTECHDS

A method for the isolation and purification of proteins (CM-cellulase, hemicellulase, protease or antibody) or cells in aqueous two-phase systems (ATPS) is new and involves combining a desired protein or a cell to an amphipathic or hydrophobic target protein (I) having the ability to separate in ATPS and to carry the protein or cell into 1 of the phases, and subjecting the fusion protein or cell carrying (I) to an ATPS separation. Also claimed are: a fusion protein (II) having (I) fused to a desired protein; a recombinant organism producing (II); a recombinant DNA with a DNA molecule encoding (II); preparation of (II) by transforming an organism with DNA enabling expression of (I) and recovering the protein from the organism culture; and separating hydrophobins or hydrophobin-like proteins in ATPSs by mixing solutions containing the hydrophobin-like protein with phase forming chemicals and carrying out ATPS separation. The method is used to isolate and purify proteins or cells in ATPSs. In an example, Saccharomyces cerevisiae VTT-C-99315 and H2155 (plasmid pYES2) were cultivated were used for experiments. (109pp)

L68 ANSWER 15 OF 21 USPATFULL on STN

ACCESSION NUMBER: 1999:75157 USPATFULL

TITLE: Method for preparing hydrophobic fumed silica

Griffith, Phillip Joseph, Llandough, United Kingdom Herron, William, South Glamorgan, United Kingdom Harkness, Brian Robert, Vale of Glamorgan, United

Kingdom

Taylor, Rosemary Margaret, Vale of Glamorgan, United

Kingdom

Wilson, David James, South Glamorgan, United Kingdom Dow Corning Corporation, Midland, MI, United States

(U.S. corporation)

NUMBER KIND DATE

NTI TM

TENT INFORMATION:

US 5919298 US 1998-5852 19990706 19980112 (9)

PLICATION INFO.: CUMENT TYPE:

LE SEGMENT:

Utility Granted

IMARY EXAMINER:

Brunsman, David Boley, William F.

GAL REPRESENTATIVE: MBER OF CLAIMS:

EMPLARY CLAIM: NE COUNT:

727

S INDEXING IS AVAILABLE FOR THIS PATENT.

A method for the preparation of hydrophobic fumed silicas which are useful, for example, as reinforcing fillers in rubber compositions. The method comprises two steps, where in the first step an aqueous suspension of fumed silica is contacted with an organosilicon compound in the presence of a catalytic amount of an acid to effect hydrophobing of the fumed silica. In the preferred method the first step is conducted in the presence of a water miscible organic solvent which facilitates hydrophobing of the fumed silica with the organosilicon compound and the fumed silica has a BET surface area greater than 50 m.sup.2 /g. In the second step the aqueous suspension of the fumed silica is contacted with a water-immiscible organic solvent at a solvent to silica weight ratio greater than 0.1:1 to effect separation of the hydrophobic fumed silica from the aqueous phase. In a preferred process the hydrophobic fumed silica has a surface area within a range of about 100 m.sup.2 /g to 750 m.sup.2 /g.

### S INDEXING IS AVAILABLE FOR THIS PATENT.

8 ANSWER 16 OF 21 USPATFULL on STN

CESSION NUMBER:

1999:63133 USPATFULL

TLE:VENTOR(S): Method of preparing hydrophobic precipitated silica Griffith, Phillip J., Llandough, United Kingdom Harkness, Brian R., Cowbridge, United Kingdom Herron, William, Cowbridge, United Kingdom Taylor, Rosemary M., Barry, United Kingdom Wilson, David J., Penarth, United Kingdom

TENT ASSIGNEE(S):

Dow Corning Corporation, Midland, MI, United States

(U.S. corporation)

NUMBER KIND DATE -----

TENT INFORMATION:

GAL REPRESENTATIVE:

US 5908660

19990601 19970903

PLICATION INFO.: CUMENT TYPE:

US 1997-923073 Utility Granted

LE SEGMENT: IMARY EXAMINER:

Cameron, Erma Boley, William F.

MBER OF CLAIMS: EMPLARY CLAIM:

20 1 446

VE COUNT:

S INDEXING IS AVAILABLE FOR THIS PATENT.

A method for the preparation of hydrophobic precipitated silicas which are useful, for example, as reinforcing fillers in rubber compositions. The method comprises two steps, where in the first step an aqueous suspension of precipitated silica is contacted with an organosilicon compound in the presence of a catalytic amount of an acid to effect hydrophobing of the precipitated silica. In the second step the aqueous suspension of the hydrophobic precipitated silica is contacted with a water-immiscible organic solvent at a solvent to silica weight ratio greater than 5:1 to effect separation of the hydrophobic precipitated silica from the aqueous phase.

### S INDEXING IS AVAILABLE FOR THIS PATENT.

ANSWER 17 OF 21 USPATFULL on STN 95:1293 USPATFULL

CESSION NUMBER:

TLE: Non-aqueous liquid cleaning products comprising

polyalkoxylated derivatives of castor oil ricinoleic

acid and analogous fatty alcohols

Houghton, Mark P., Rotterdam, Netherlands

Verburg, Charles C., Vlaardingen, Netherlands

Lever Brothers Company, Division of Conopco, Inc., New

York, NY, United States (U.S. corporation)

NUMBER KIND DATE

TENT INFORMATION: US 5378387 19950103

US 1993-71436 19930601 (8)

NUMBER DATE

IORITY INFORMATION: EP 1992-201565 19920602

CUMENT TYPE: Utility

LE SEGMENT: Granted
IMARY EXAMINER: Lieberman, Paul

SISTANT EXAMINER: Hertzog, A.
GAL REPRESENTATIVE: Koatz, Ronald A.

MBER OF CLAIMS: 5
EMPLARY CLAIM: 1,4
NE COUNT: 700

VENTOR(S):

TENT ASSIGNEE(S):

PLICATION INFO.:

S INDEXING IS AVAILABLE FOR THIS PATENT.

Substantially non-aqueous liquid cleaning compositions comprising a non-aqueous liquid phase that comprises a polyalkoxylated castor oil derivative and/or polyalkoxylated derivatives of ricinoleic acid (set forth as formula (III)) and/or polyalkoxylated derivatives of analogous fatty alcohols (set forth as formula (IV)).

### S INDEXING IS AVAILABLE FOR THIS PATENT.

8 ANSWER 18 OF 21 USPATFULL on STN

CESSION NUMBER: 82:39765 USPATFULL

TLE: Method for producing hydrophobic reinforcing silica

fillers and fillers obtained thereby

VENTOR(S): Lutz, Michael A., Midland, MI, United States

TENT ASSIGNEE(S): Dow Corning Corporation, Midland, MI, United States

(U.S. corporation)

TENT INFORMATION: US 4344800 19820817
PLICATION INFO:: US 1980-156002 19800603 (6)

CUMENT TYPE: Utility
LE SEGMENT: Granted
IMARY EXAMINER: Dees, Carl F.
GAL REPRESENTATIVE: Rakoczy, Richard E.
MBER OF CLAIMS: 84

EMPLARY CLAIM: 1,43
NE COUNT: 2072

S INDEXING IS AVAILABLE FOR THIS PATENT.

Hydrophobic reinforcing silica fillers for silicone rubber are produced by the steps of mixing an alkyl silicate, preferably methyl orthosilicate, at least 70% of one-half of the stoichiometric amount of water required to hydrolyze the alkoxy radicals present in the alkyl silicate, an alcohol and at least 0.05 moles (per mole of theoretical SiO.sub.2 units present in the alkyl silicate) of a hydrophobe agent such as hexamethyldisilazane together in the presence of a basic catalyst, said hydrophobe agent being added prior to the gelation of the mixture, and aging the mixture to obtain a composition containing a hydrophobic reinforcing silica filler for silicone rubber. Preferably, the hydrophobe agent is added prior to or concurrently with the addition of the alkyl silicate. Vulcanized silicone rubbers possessing tensile strengths in excess of 12.4 megapascals and tear strengths of greater

S INDEXING IS AVAILABLE FOR THIS PATENT.

```
ANSWER 19 OF 21 DGENE COPYRIGHT 2004 THOMSON DERWENT on STN
CESSION NUMBER: AAA95429 DNA
                                    DGENE
              Isolation and purification of proteins or cells in
                aqueous two-phase systems,
                comprises combining a desired protein or a cell to a
                targeting protein capable of isolating the protein or cell
                into one of the phases -
VENTOR:
                Penttilae M; Nakari-Setaelae T; Fagerstroem R; Selber K; Kula
                M; Linder M; Tjerneld F
TENT ASSIGNEE:
                (VALW) VALTION TEKNILLINEN TUTKIMUSKESKUS.
TENT INFO:
                WO 2000058342 A1 20001005
PLICATION INFO: WO 2000-FI249
                                 20000324
IORITY INFO:
               FI 1999-667
                                 19990325
                FI 1999-1782
                                 19990820
CUMENT TYPE:
                Patent
NGUAGE:
                English
HER SOURCE:
                2000-686858 [67]
                S. commune SC3 coding sequence PCR primer #2.
SCRIPTION:
    AAA95429 DNA
                        DGENE
    The present invention is related to a novel method for separating,
    purifying and isolating proteins and cells. This involves the use of
    liquid-liquid extraction in an aqueous two-
    phase system (ATPS) which partitions molecules by fusing them to
    targeting proteins which then carry the molecules of interest into one of
    the phases. The present sequence is a PCR primer which was used to
    demonstrate the method of the invention. The method is useful in also
    useful in the identification of nucleic acid sequences in expression
    library screening.
   ANSWER 20 OF 21 DGENE COPYRIGHT 2004 THOMSON DERWENT on STN
CCESSION NUMBER: AAA95428 DNA
                                    DGENE
TLE:
                Isolation and purification of proteins or cells in
                aqueous two-phase systems,
                comprises combining a desired protein or a cell to a
                targeting protein capable of isolating the protein or cell
                into one of the phases -
IVENTOR:
                Penttilae M; Nakari-Setaelae T; Fagerstroem R; Selber K; Kula
                M; Linder M; Tjerneld F
                (VALW) VALTION TEKNILLINEN TUTKIMUSKESKUS.
ATENT ASSIGNEE:
ATENT INFO:
                WO 2000058342 A1 20001005
                                                        109p
PPLICATION INFO: WO 2000-F1249
                                 20000324
                FI 1999-667
RIORITY INFO:
                                 19990325
                FI 1999-1782
                                 19990820
CUMENT TYPE:
                Patent
ANGUAGE:
                English
THER SOURCE:
                2000-686858 [67]
SCRIPTION:
                S. commune SC3 coding sequence PCR primer #1.
    AAA95428 DNA
                        DGENE
    The present invention is related to a novel method for separating,
    purifying and isolating proteins and cells. This involves the use of
    liquid-liquid extraction in an aqueous two-
    phase system (ATPS) which partitions molecules by fusing them to
    targeting proteins which then carry the molecules of interest into one of
    the phases. The present sequence is a PCR primer which was used to
    demonstrate the method of the invention. The method is useful in also
    useful in the identification of nucleic acid sequences in expression
    library screening.
    ANSWER 21 OF 21 DGENE COPYRIGHT 2004 THOMSON DERWENT on STN
CCESSION NUMBER: AAA95427 DNA
                                    DGENE
TLE:
                Isolation and purification of proteins or cells in
```

aqueous two-phase systems,

comprises combining a desired protein or a cell to a

targeting protein capable of isolating the protein or cell

Penttilae M; Nakari-Setaelae T; Fagerstroem R; Selber K; Kula

into one of the phases -

M; Linder M; Tjerneld F PATENT ASSIGNEE:

(VALW) VALTION TEKNILLINEN TUTKIMUSKESKUS.

PATENT INFO: WO 2000058342 A1 20001005 APPLICATION INFO: WO 2000-F1249 20000324 FI 1999-667 PRIORITY INFO: 19990325

FI 1999-1782 19990820

OCUMENT TYPE: Patent LANGUAGE: English

INVENTOR:

lΝ

łΒ

2000-686858 [67]

THER SOURCE: DESCRIPTION:

S. commune SC3 coding sequence. AAA95427 DNA

DGENE

The present invention is related to a novel method for separating, purifying and isolating proteins and cells. This involves the use of liquid-liquid extraction in an aqueous two-

phase system (ATPS) which partitions molecules by fusing them to targeting proteins which then carry the molecules of interest into one of the phases. The present sequence was used in a fusion molecule to demonstrate the method of the invention. The method is useful in also useful in the identification of nucleic acid sequences in expression library screening.

# **WEST Search History**

Hide Items Restore Clear Cancel

ATE: Wednesday, May 26, 2004

ide?	<u>Set</u> Name	Query	<u>Hit</u> Count
	DB=PC	GPB,USPT,USOC,EPAB,JPAB,DWPI,TDBD; THES=ASSIGNEE; PLUR=YES; OP=ADJ	•
	L23	(hfbI or hfbII or srhI or sc3) and (aqueous same two adj phase)	4
	L22	(hfb\$ or srhI or sc3) and (aqueous same two adj phase)	17
	L21	(hfb\$ or srhI or sc3) and (aqueous adj two adj phase)	1
	L20	(hfb\$ or srhI or sc3) and (aqueous adj two adj phase or ATPS)	215
	L19	(hfb\$ or srhI or sc3) and (aqueous adj two adj phase or ATPS)	215
	L18	(hfb\$ or srhI or sc3) and (aqueous same two adj phase or ATPS)	231
	L17	(hfb\$ or srhI or sc3) and (aqueous same two same phase or ATPS)	260
П	L16	(hfb\$ or srhI) and aqueous same phase	179
	L15	(hfb\$ or srhI) and hydrophobin and aqueous same phase	. 1
	L14	(hfb\$ or srhI) same hydrophobin and aqueous same phase	0
-	DB=PC	GPB,USPT,EPAB,DWPI,TDBD; THES=ASSIGNEE; PLUR=YES; OP=ADJ	
	L13	(hfb\$ or srhI) same hydrophobin and aqueous same phase	0
	L12	sc3 same hydrophobin and aqueous same phase	0
	L11	sc3 same hydrophobin and two same phase	0
	L10	sc3 same hydrophobin	5
Г	L9	(hydrophobin or amphipath\$ or HFBI or HFBII or SRHI ) same aqueous same two adj phase	11
	L8	(hydrophobin or surfactant or amphipath\$ or HFBI or HFBII or SRHI ) same aqueous same two adj phase	620
	L7	(hydrophobin or surfactant or amphipath\$ orHFBI or HFBII or SRHI ) same aqueous same two adj phase	611
	L6	(hydrophobin or surfactant or amphipath\$) and aqueous same two adj phase	3799
	L5	hydrophobin and aqueous same two adj phase	1
Г	L4	hydrophobin? and aqueous same two adj phase	6
	L3	hydrophobin\$ and aqueous same two adj phase	6
	L2	hydrophobin\$ and aqueous adj two adj phase	1
	L1	hydrophobin\$	764

ND OF SEARCH HISTORY

## **Hit List**

Clear Generate Collection Print Fwd Refs Bkwd Refs Generate OACS

**Search Results -** Record(s) 1 through 6 of 6 returned.

1. Document ID: US 20020131147 A1

ing default format because multiple data bases are involved.

L4: Entry 1 of 6

File: PGPB

Sep 19, 2002

PUB-DOCUMENT-NUMBER: 20020131147

PUB-FILING-TYPE: new

CUMENT-IDENTIFIER: US 20020131147 A1

TLE: Electrophoretic medium and process for the production thereof

BLICATION-DATE: September 19, 2002

VENTOR-INFORMATION:

ME CITY STATE COUNTRY RULE-47 Arlington olini, Richard J. JR. MA US

ller, David D. Billerica US MA miskey, Barrett Cambridge US MA

-CL-CURRENT: <u>359</u>/296; 359/290

Full Title Citation	on Front Review	Classification	Date	Reference	Sequences	Attachments	Claims	KWIC	Draww Desc	Image
					2					
······································	***************************************	W		• • • • • • • • • • • • • • • • • • • •	**************************************			*******************	**************************************	*******************

☐ 2. Document ID: US 5919298 A

L4: Entry 2 of 6 File: USPT Jul 6, 1999

-PAT-NO: 5919298

CUMENT-IDENTIFIER: US 5919298 A

TLE: Method for preparing hydrophobic fumed silica

FE-ISSUED: July 6, 1999

VENTOR-INFORMATION:

ME	CITY	STATE	ZIP CODE	COUNTRY
iffith; Phillip Joseph	Llandough			GB
rron; William	South Glamorgan			GB
rkness; Brian Robert	Vale of Glamorgan			GB
ylor; Rosemary Margaret	Vale of Glamorgan			GB
lson; David James	South Glamorgan			GB

L-CURRENT: 106/490; 423/336, 423/337

		•
Full   Title   Citation   Front   Review   Classification   D	ate Reference 3-Highwey Anxientes	Claims KWC Draw Desc Image
3. Document ID: US 5908660 A		
Entry 3 of 6	File: USPT	Jun 1, 1999
I-NO: 5908660 ENT-IDENTIFIER: US 5908660 A		

ee image for Certificate of Correction \*\*

E: Method of preparing hydrophobic precipitated silica

-ISSUED: June 1, 1999

NTOR-INFORMATION:

	CITY	STATE	ZIP CODE	COUNTRY
fith; Phillip J.	Llandough			GB
mess; Brian R.	Cowbridge			GB
on; William	Cowbridge			GB
or; Rosemary M.	Barry			GB
on; David J.	Penarth			GB

L-CURRENT: 427/220; 106/490, 427/221, 427/443.2

Full Title	Citation Front	Review	Classification	Date	Reference		112	Claims	KWIC	Draw, Des	c Image
-											
***************************************	***************************************						***************************************	<del>,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,,</del>			
☐ 4. Do	cument ID:	US 5378	387 A								
4: Entry 4	of 6		•		Fil	e: USPT				Jan 3	, 1995

AT-NO: 5378387

MENT-IDENTIFIER: US 5378387 A

E: Non-aqueous liquid cleaning products comprising polyalkoxylated derivatives of castor ricinoleic acid and analogous fatty alcohols

-ISSUED: January 3, 1995

NTOR-INFORMATION:

€	CITY	STATE	ZIP CODE	COUNTRY
ghton; Mark P.	Rotterdam			NL
ourg; Charles C.	Vlaardingen			NL

L-CURRENT: 510/161; 510/221, 510/235, 510/304, 510/312, 510/338, 510/356, 510/413, 510/437

Full	Title	Citation	Front	Review	Classification	Date	Reference	r participation	Claims	KWC	Drawi Desc	Image

5. Document ID: US 4344800 A

1: Entry 5 of 6

File: USPT

Aug 17, 1982

AT-NO: 4344800

MENT-IDENTIFIER: US 4344800 A

ee image for Certificate of Correction \*\*

E: Method for producing hydrophobic reinforcing silica fillers and fillers obtained thereby

-ISSUED: August 17, 1982

NTOR-INFORMATION:

CITY

Full Title Citation Front Review Classification Date Reference

STATE

ZIP CODE

COUNTRY

Claims KWC Draw Desc Image

; Michael A.

Midland

ΜI

L-CURRENT: 106/481; 106/490, 502/158, 502/200, 524/588, 524/860

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	ID NO	61 4001 A 337	2000050242	1 411 20002 7 (2)	Δ FP 1163260 Δ	1 310 000101501

A, KR 2001108400 A, CN 1357005 A, JP 2002543766 W

4: Entry 6 of 6

File: DWPI

Oct 31, 2003

ENT-ACC-NO: 2000-686858

ENT-WEEK: 200380

RIGHT 2004 DERWENT INFORMATION LTD

E: Isolation and purification of proteins or cells in <u>aqueous two-phase</u> systems, comprises ining a desired protein or a cell to a targeting protein capable of isolating the protein ell into one of the phases

NTOR: FAGERSTROEM, R; KULA, M ; LINDER, M ; NAKARI-SETAELAE, T ; PENTTILAE, M ; SELBER, K ; NELD, F ; FAGERSTROM, R ; NAKARI-SETALA, T ; PENTTILA, M

RITY-DATA: 1999FI-0001782 (August 20, 1999), 1999FI-0000667 (March 25, 1999)

#### NT-FAMILY:

NO	PUB-DATE .	LANGUAGE	PAGES	MAIN-IPC
14891 A	October 31, 2003		000	C07K001/14
00058342 A1	October 5, 2000	E	109	C07K001/14
00035621 A	October 16, 2000		000	C07K001/14
163260 A1	December 19, 2001	E	000	C07K001/14
00104534 A	November 26, 2001	*	000	C07K000/00
001108400 A	December 7, 2001		000	C07K001/20
357005 A	July 3, 2002		000	C07K001/14
002543766 W	December 24, 2002		112	C12N015/09

CL (IPC): <u>B01</u> <u>D</u> <u>17/025</u>; <u>B01</u> <u>D</u> <u>17/038</u>; <u>C07</u> <u>K</u> <u>0/00</u>; <u>C07</u> <u>K</u> <u>1/14</u>; <u>C07</u> <u>K</u> <u>1/20</u>; <u>C07</u> <u>K</u> <u>14/37</u>; <u>C07</u> <u>/00</u>; <u>C12</u> <u>N</u> <u>1/15</u>; <u>C12</u> <u>N</u> <u>1/19</u>; <u>C12</u> <u>N</u> <u>1/21</u>; <u>C12</u> <u>N</u> <u>5/10</u>; <u>C12</u> <u>N</u> <u>9/24</u>; <u>C12</u> <u>N</u> <u>15/09</u>; <u>C12</u> <u>N</u> <u>15/62</u>; <u>P</u> <u>21/02</u>; <u>C12</u> <u>R</u> <u>1:885</u>

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HASE	1312595
PHASES	277510
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	Eur J Bioche	nd biochemical chara em: 1996 Jan 15;235(1-2 :337 [PubMed - indexed	2):248-55.	e Trichoderma r	eesei hydrophob	in HFBI.
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	Hydrophobin gene srh1, expressed during sporulation of the biocontrol Trichoderma harzianum.  Curr Genet. 1997 Sep;32(3):225-30.  PMID: 9339348 [PubMed - indexed for MEDLINE]	ol agent		
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	Localization and cell surface anchoring of the Saccharomyces cerevis protein Flo1p. J Bacteriol. 1997 Aug;179(15):4929-36. PMID: 9244284 [PubMed - indexed for MEDLINE]	siae flocculation		
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